{Exhibit 78}

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ELISA

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Edited by
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- the incubation and washing stages
- the addition of enzyme-labelled antigen or antibody
- the addition of specific substrate
- the measurement and interpretation of results.

Although the theory of ELISA is elegantly simple, there are many problems which may be encountered at the various stages. This is true whether an established procedure is being followed or a new assay is being set up. This chapter highlights these problems – some of which are more obvious than others – and suggests ways in which these may be avoided.

The choice of solid phase

Many different types of solid phase have been used for ELISAs, including particulate materials such as cellulose [1], polyacrylamide [7] and agarose [2] to which antigen or antibody is covalently bound. These have the disadvantage that, with the exception of ferritin-containing particles which are magnetic, the washing steps involve centrifugation. The use of such carriers as microtitre plates, tubes, macrobeads or cuvettes avoids this problem. All these solid phases are available in a variety of materials – eg polystyrene, polyvinyl, polycarbonate and nylon. The material itself may be either treated (eg gamma irradiated) or untreated. The most widely used carrier is the 96-well microtitre plate; the majority of instrumentation is available for this carrier.

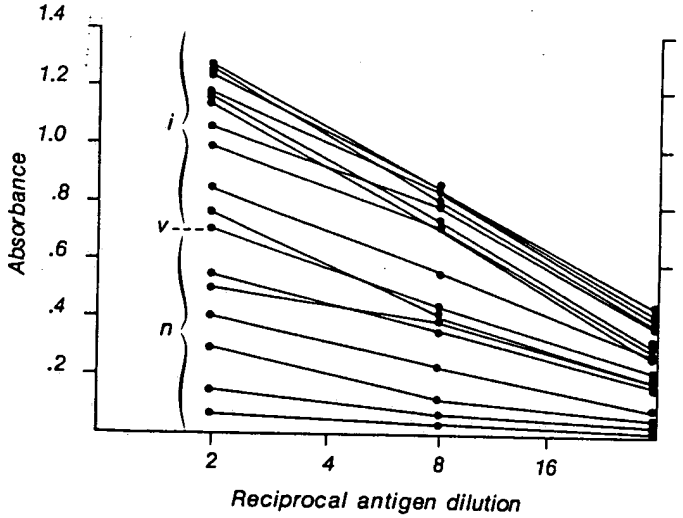

Figure 3 Double antibody sandwich for detection of rotavirus antigen. A comparison of different microtitration plates – polystyrene, irradiated (i) and non-irradiated (n), and polyvinyl (v)